Antioxidant Supplements, Genetics and Chemotherapy Outcomes

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Abstract: Cancer patients report widespread use of antioxidant supplements during chemotherapy, despite recommendations by the American Institute for Cancer Research and others that supplements should not be used during treatment. These guidelines are based upon the fact that numerous chemotherapeutic agents, as well as radiation therapy, exert their cytotoxic effects by generation of reactive oxygen species (ROS), which cause massive damage to DNA and proteins and trigger apoptosis, resulting in tumor and normal cell death. Thus, there is the concern that antioxidants may block the ROS-generated effects of therapy on tumor cells. There are no data based on sound epidemiological or clinical studies to support this hypothesis, however. In fact, some experimental studies have shown that antioxidants may potentiate the effects of chemotherapeutic drugs, while also lessening treatment-related toxicities. In this report, we review the literature regarding chemotherapy and radiation therapy as sources of oxidative stress, and present the current data regarding effects of antioxidant supplement use on normal and cancer cells. The role of antioxidant supplements, as well as the role of genetic variants in oxidative stress genes, in relation to cancer treatment toxicities and survival are discussed.

Keywords: Antioxidant supplements, reactive oxygen species, polymorphisms, chemotherapy, toxicity, survival.

INTRODUCTION

Although there is a widespread use of antioxidant supplements by cancer patients during therapy, there are no clinical guidelines for physicians to use to advise their patients on complementary approaches to therapy. Nonetheless, because of the potential for antioxidants to block reactive oxygen species (ROS) generated by alkylating agents and anthracyclines, as well as taxanes, it has been suggested that physicians instruct their patients to discontinue all non-conventional nutritional supplementation during the course of anticancer therapy [1]. In fact, the American Institute for Cancer Research recommended in 2003, that "taking dietary supplements containing levels of nutrients with antioxidant properties much greater than the dietary reference intakes (DRI) is not recommended during chemotherapy, because higher levels may have adverse effects and interfere with efficacy of treatment" [2,3]. Because there is potential for antioxidants to reduce toxicities associated with chemotherapeutic regimens, as well as to actually maximize the effects of treatment agents, there is a need to evaluate the effects of use of antioxidant supplements during cancer therapy.

In this report, we review the literature regarding chemotherapy and radiation therapy as sources of oxidative stress, and present the current data regarding effects of antioxidants on normal and cancer cells. Furthermore, because the effects of ROS on normal and tumor tissue may also be modified by endogenous oxidant and antioxidant capabilities, the role of antioxidant supplements, as well as the role of genetic variants in oxidative stress genes, in relation to cancer treatment toxicities and survival are discussed.

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1. USE OF ANTIOXIDANT SUPPLEMENTS BY BREAST CANCER PATIENTS

Use of complementary medicine, particularly antioxidant supplements, is widespread among cancer patients. In the Women's Healthy Eating and Living (WHEL) Study [4], a national, multi-center study of > 3000 women with breast cancer, 59% of women were reported taking multivitamins and more than 40% used vitamin C and E supplements. In the Western New York Exposures and Breast Cancer Study (WEB Study), more than 1000 cases and 2000 controls were queried regarding supplement use [5]. In that study, 67% of cases and only 39% of controls reported supplement use during the period following diagnosis for cases and reference date for controls. 42% of cases specifically reported taking antioxidants, while only 16% of controls. Use of large doses of antioxidants is also not uncommon. In the WHEL study [4], more than 1/4th of the women reported taking megadoses of Vitamin C (> 1000mg (12.5 times the RDA)), and in a study of 500 women with breast and gynecological malignancies conducted at MD Anderson Cancer Center [6], it was found that 29% of women used megadoses of vitamin E, with 12% using megadoses of Vitamin C and 10% megadoses of Vitamin B. The term mega-vitamin and/or mineral was defined as greater than or equal to 50 times the recommended daily allowance in this study.

Although many clinicians advise their patients not to take antioxidants during treatment [1], it appears that patients do not routinely report use of supplements to their physicians. A general survey reported that 61% of patients did not disclose the use of complementary medicines to their physicians [7], and another study found that over 30% of breast cancer patients used megavitamins without discussing their use with their doctor [8]. Interestingly, of women queried in the MD Anderson study, only 28% considered supplements to be medications that should be reported to their health care practitioners [6]. Because of the widespread use of supplements by cancer patients, and the lack of disclosure of supplement use to physicians, there is a clear need for well-

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designed studies to determine the potential effects of antioxidant supplement use during chemotherapy on cancer outcomes.

2. CHEMOTHERAPY AND OXIDATIVE STRESS

2.1. Generation of ROS by Chemotherapy

Experimental and clinical studies have shown that a major mechanism for the cytotoxic activity of numerous chemotherapeutic agents is through increased formation of ROS, including hydroxyl radicals (OH•), hydrogen peroxide (H_2O_2) and superoxide anions ($\bullet O^2$-) [9-12]. A number of clinical studies have shown that patients treated with a wide range of cytotoxic agents, but particularly cyclophosphamide and adriamycin, have marked increases in lipid peroxidation products [13-18], as well as decreases in vitamin E concentrations [16] after treatment.

The chemotherapeutic agents cyclophosphamide, doxorubicin (adriamycin) and paclitaxel, now commonly used for treatment of breast cancer, have all been shown to increase lipid peroxidation and generation of ROS [19]. Numerous studies have noted that administration to rats of cyclophosphamide or its metabolite, acrolein, results in an increase in lipid peroxidation products, such as malondialdehyde [20-22]. Cyclophosphamide exposure also results in concomitant decreases in glutathione [22] and the endogenous antioxidants, superoxide dismutase and glutathione peroxidase [18]. The mechanism of cyclophosphamide's tumor cell kill by ROS is further demonstrated by the rodent data showing that the lung injury associated with treatment with cyclophosphamide is due to its ability to generate free radicals [17,21].

Doxorubicin, like other anthracyclines, results in the formation of quinone-mediated free radicals, which have the capacity to cause oxidative damage and cytotoxicity. The fact that the drug's tumor cell-killing mechanism is, in part, through oxidative stress is demonstrated by data showing that adriamycin's cardiotoxicity is a result of the production of ROS [23]. These ROS generated by adriamycin are presumably acting on tumor cells as well. Cellular oxidoreductases reduce adriamycin to a semiquinone radical that is subsequently reoxidized by oxygen to a superoxide anion and the parent quinone [12,24,25]. Superoxide anions can dismutate to form hydrogen peroxide and/or react with nitric oxide to form peroxynitrite; thus, while use of antioxidant supplements may impact levels of ROS, cellular levels of superoxide dismutase, catalase and glutathione peroxidase are also important in determining amounts of these ROS present. Lipid peroxides resulting from doxorubicin can further break down to yield hydroxyalkenals, which are substrates for glutathione-conjugating isozymes [26], indicating the potential importance of the glutathione-S transferases in mediating therapeutic outcomes.

Taxols interfere with microtubular disassembly, ultimately resulting in DNA fragmentations and features of apoptosis [27]. Through signaling pathways, paclitaxel leads to increased levels of Bax, activation of caspase proteins, and ultimately to induction of mitochondrial ROS production and mitochondrial permeability, resulting in generation of high levels of ROS [28], as further described below. In a study of lymphoma cell lines, it was noted that treatment with paclitaxel resulted in increased generation of ROS, and that this increase was suppressed by antioxidants, including endogenous (catalase) and exogenous (ascorbic acid) forms [29]. Thus, all three drugs currently used in the treatment of breast cancer result in potentially cytotoxic ROS, and there are *in vitro* data indicating that chemotherapeutic agents interact with antioxidants in their effects on cell death.

2.2. ROS, Mitochondria and Apoptosis

One important mechanism for treatment efficacy and tumor kill is programmed cell death, or apoptosis [30], and disruption of apoptotic programs can reduce treatment sensitivity [31]. Specific chemotherapeutic agents used to treat breast cancer, cyclophosphamide and doxorubicin [32] and paclitaxel [28], have been shown to induce apoptosis in malignant cells. Because apoptosis is a regulated process, factors that make cells more or less susceptible to apoptosis are likely to affect their sensitivity to a wide range of chemotherapeutic agents.

It is widely held that chemotherapeutic agents induce mitochondrial changes and apoptosis through mechanisms associated with ROS production [33,34]. As reviewed by Mignotte [35], it has been well documented that oxidative stress provokes cell death as a result of massive cellular damage associated with lipid peroxidation and alterations of proteins and nucleic acids, although other pathways are likely to be important as well. Apoptosis occurs when, through a pathway of signaling, the mitochondrial membrane becomes permeable [31,36,37]. Mitochondria are the main site for ROS generation and are thought to be a major intracellular target for oxidative damage [38]. Anticancer agents can cause mitochondrial permeability through enhanced generation of ROS, and once the mitochondrial membrane barrier function is lost, a number of other factors contribute to cell death. While ROS, among other factors, induce or facilitate mitochondrial permeability, glutathione and antioxidant enzymes such as manganese superoxide dismutase (MnSOD), catalase (CAT) and glutathione peroxidase (GPX1) inhibit it [37]. In fact, experimental results indicate that MnSOD prevents the disruption of mitochondrial membrane potential [39]. It was shown that inhibition of SOD caused accumulation of superoxide radicals, leading to free-radical-mediated damage to mitochondrial membranes and apoptosis of cancer cells [40], and in a commentary on the study of MnSOD and apoptosis of cancer cells, Cleveland and Kastan [41] suggested that a promising way of treating some cancers could be by increasing levels of ROS and inhibition of SOD. All of these data clearly indicate that 1) ROS are important in the cell-killing effects of chemotherapeutic agents and 2) antioxidants (both supplements and endogenous forms) may impact the ultimate effects of ROS on both normal and tumor cells.

3. ANTIOXIDANTS AND CANCER THERAPY

3.1. Vitamin Supplements

The potential role of antioxidant supplements in cancer therapy outcomes has been investigated for a number of years through research in cell lines, in animals, and in small patient populations. The data resulting from these studies

are, for the most part, conflicting, and supplement use during treatment remains a controversial area. Perhaps, at the crux of these conflicting data is the growing recognition that antioxidants can also act as pro-oxidants, depending upon the cell type (normal *vs*. neoplastic), dose, and general oxidative state [42]. The two main schools of thought regarding antioxidant supplement use during therapy, are described below.

3.1.1. Antioxidants Block Therapeutic Efficacy

Numerous scientists argue that antioxidant supplements may interfere with the effects of ROS on tumor cell DNA and membranes [1], as well as reduce the anti-apoptotic breakdown of tumor cells [43]. Because numerous anticancer drugs kill cancer cells by triggering apoptosis, there is concern that antioxidants can deplete ROS and interfere with this apoptotic pathway, as reviewed by Salganik [44]. Evidence for this potential mechanism is discussed in the preceding section and consists of experimental work performed in numerous cell lines with different chemotherapy agents. For example, Salganik *et al*. showed that therapy-induced apoptosis was accompanied by an increase in ROS generation, and that alpha-tocopherol inhibited ROS generation as well as apoptosis of cancer cells [44]. However, there have been no well-designed studies that have addressed this issue in humans, and few small studies that have provided sufficient data for this hypothesis to be supported. Nonetheless, it is still recommended that clinicians advise their patients not to take antioxidant supplements during treatment [1-3].

3.1.2. Antioxidants Enhance Treatment Efficacy

Some investigators suggest that antioxidant supplements may increase efficacy of treatments while reducing therapy-related side effects [45,46]. As summarized by Prasad [47], this is based on the hypothesis that the mechanism of action of dietary antioxidants on cancer cells may be different than that observed for normal cells. Prasad commented that these differences may be due to a number of mechanisms, including: 1) differential accumulation of antioxidants by normal and cancer cells, as demonstrated in animals [48] and in leukemia patients [45]; 2) differing requirements for oxidative stress in tumor *vs*. normal cells, with reduction of ROS in cancer cells possibly leading to proliferation inhibition and increased apoptosis [49]; 3) differences in transcription factor activities that affect changes in the pattern of gene expression [50], with some pathways linked to enhanced cell survival and others to cell death, depending upon cell type. It has also been suggested that excessive oxidative stress may slow down the cell cycle, resulting in slower proliferation rates [34]. Because chemotherapeutic agents are most cytotoxic to cells that are rapidly proliferating, it has been hypothesized that reduction of ROS by antioxidants could thereby enhance the effects of chemotherapy [34]. Regardless of the specific pathway, it is clear from experimental data that, in many situations, antioxidants given along with chemotherapy actually enhance tumor cell kill [9,31,34,51]. In fact, Rustum *et al*. have recently shown that selenium significantly reduced toxicity associated with a number of chemotherapeutic agents, while also enhancing efficacy and increasing cure rates among nude mice xenografts [52].

3.2. Clinical Studies of Antioxidant Use, Treatment Toxicities and Survival

Despite these compelling experimental data, there are few examples of such associations among cancer patients, with most reports based on very small sample sizes [53]. For example, 20 patients with oral cancer who received supplemental dietary beta carotene experienced less severe mucositis when treated with radiation therapy and chemotherapy than those not receiving a supplement, although there were no differences in recurrence rates [54]. In a lung cancer study of 18 patients treated with cyclophosphamide, adriamycin and vincristine, supplementation with antioxidants resulted in longer survival than those not receiving supplements [55]. Patients receiving supplements also were able to tolerate chemotherapy better. Recently, it was shown that toxicities were reduced in a group of patients treated with cisplatin (n=27), who received 300 mg/day of vitamin E [56]. In one larger study with breast cancer patients in British Columbia [57], women with non-metastatic breast cancer were treated with megadoses of varying amounts of beta carotene, niacin B3, vitamin C, selenium, coenzyme Q10 and zinc. There was no consistency in whether or not women received chemotherapy or radiation therapy or both, and supplements were started up to 6 months after diagnosis. Although not statistically significant, women receiving supplements had poorer survival than those who were not treated with vitamins. This study had several design limitations including small sample size, heterogeneous population, varied treatments and varying doses of supplements at differing time points. More recently, 49 women receiving treatment for breast cancer were queried regarding supplement use [58]. In this small study, women taking supplements, particularly vitamin E, had less reduction in neutrophil counts than women not taking supplements. Furthermore, in a study of 133 patients undergoing stem cell transplantation for leukemia, mucositis was less severe among patients who were taking multivitamins prior to transplantation [59].

Although some research is being conducted to evaluate the effects of diet and supplement use on breast cancer survival, such as the WHEL study, these studies address supplement use after breast cancer treatment and do not have the ability to evaluate the effects of antioxidant use during treatment on both toxicity and survival. Furthermore, while some studies report on high doses of antioxidants used in experimental conditions, they do not provide information on the effects of lower doses that are taken by patients in non-controlled settings. Prasad *et al*. argue that, while at high doses, antioxidants may enhance the effects of chemotherapeutic agents, low dose vitamins may block efficacy to some extent [60]. Clearly, large studies are needed to clarify these issues, with patients on standardized chemotherapy and data on supplement use at entry and at completion of chemotherapy, as well as additional other clinical and lifestyle factors that could impact relationships between supplement use and disease-free survival.

3.3. Endogenous Influences on Oxidative Stress

While ROS, among other factors, induce or facilitate mitochondrial permeability, glutathione and antioxidant enzymes such as MnSOD, CAT and GPX1 inhibit it [37].

These enzymes form the first line of defense against superoxide and hydrogen peroxide. The second line of defense against ROS is provided by enzymes such as the glutathione S-transferases α, μ, π, and θ, which remove hydrogen peroxide and other peroxides. The role of enzymes that generate ROS, such as myeloperoxidase (MPO), may also be important in total oxidative burden. Endogenous antioxidant capabilities may impact treatment-related toxicities and disease-free survival, as we have previously shown [61-63], or may modify the effects of antioxidant use on treatment outcomes. Previously, we have found that dietary antioxidants and variants in MPO [64] and CAT interact in predicting risk of breast cancer, and similar associations may exist for supplement use, genetic variants, and treatment outcomes. Several enzymes that are related to oxidative stress (MnSOD, CAT, GPX1, MPO, GSTM1, GSTT1, GSTA1 and GSTP1) are discussed below.

3.3.1. Manganese Superoxide Dismutase (MnSOD)

Manganese Superoxide Dismutase (MnSOD), which is induced with free radical challenge [65], is synthesized in the cytosol and post-transcriptionally modified for transport into the mitochondrion [66,67]. In the mitochondrion, MnSOD catalyzes the dismutation of two superoxide radicals, producing H_2O_2 and oxygen. A polymorphism in MnSOD exists in codon 16, which is located at position –9 of the mature protein and results in the incorporation of either alanine (C allele) or valine (T allele) in the mitochondrial targeting sequence. Recent experimental data indicate that the ala containing MnSOD is targeted into the mitochondria, whereas the val form of the protein is partially arrested in the inner mitochondrial membrane [68]. While one would intuitively hypothesize that the less efficient form (T) would be associated with higher levels of ROS and greater risk of cancer, it is the C polymorphism that has been associated with risk of breast [69], prostate [70], and bladder [71], but not lung [72] cancer. This increased risk with C alleles might be due to other mechanisms, such as protein-protein interactions, and subsequent disruption of MnSOD despite efficient localization to the mitochondrion. The ala-9val polymorphism has been evaluated in one small study of radiotherapy [73], with null findings. In our previous study of women receiving treatment for breast cancer, we found that, among women with MPO variants resulting in higher transcription, the MnSOD CC genotype was associated with better survival [74].

3.3.2. Catalase (CAT)

Catalase (CAT) is a heme enzyme that has a predominant role in controlling hydrogen peroxide concentration in human cells, by converting H_2O_2 into H_2O and O_2. With SOD and glutathione peroxidase (GPX), catalase constitutes a primary defense against oxidative stress and may provide resistance to the effects of chemotherapy. Indeed, chronic exposure of fibroblasts to increasing concentrations of H_2O_2 and O_2 results in the development of a stable oxidative stress-resistant phenotype characterized by increased cellular antioxidants including GPX, SOD and CAT [75]. A common polymorphism has been identified in the promoter region of the CAT gene, a -262 C→T substitution on the 5' region of the human CAT gene from the transcription start site [76]. The variant alters gene expression when incorporated upstream in a Luciferase reporter construct and is transiently transfected in HepG2 (human liver) cells and K562 (human blood cells) [76], and we have found that the T allele is associated with lower levels of red blood cell catalase activity (unpublished data). This variability in CAT activity is thought to play a role in host response to oxidative stress and, indeed, variant CAT alleles appeared to be associated with increased risk of hypertension [77] and arsenic-induced hyperkeratosis [78], conditions likely related to oxidative stress. We also found that the high activity CAT genotype was associated with reduced breast cancer risk in a large case-control study [79]. In relation to cancer survival, although we did not have adequate power in our study to detect a significant association with survival, results were suggestive that low activity CAT, in combination with high activity MPO variants, were associated with better survival [74].

3.3.3. Glutathione Peroxidases (GPX1)

Glutathione peroxidases (GPX1) are a family of enzymes that catalyze the reduction of H_2O_2 and organic hydroperoxides to water and alcohols respectively. Selenium-dependent glutathione peroxidase (GPX1) is present in the cytosol and in mitochondria [80]. GPX1 is induced under conditions of oxidative stress, protects against oxidative stress in mouse models [81], and is over expressed in cancer tissue [82-84]. In a study with two high expressing clones bearing an SFFV-GPX construct, GPX1 was found to protect against apoptotic death [85]. In another study, GPX1 expression in breast cancer cells was inversely related to ER expression, and also related to adriamycin-induced hydroxyl radical formation [86]. Particularly combined with variability in MnSOD, levels of GPX1 activity could be extremely important in cell sensitivity to treatment-generated ROS. An in-frame variable polyalanine (GCG) repeat polymorphism has been described, and the six alanine (ALA6) repeat allele also has a nucleotide substitution associated with a proline-leucine substitution [87]. Measurement of 8-hydroxydeoxyguanosine (8OHdG), a marker of oxidative damage in DNA from normal lung tissue revealed a trend of less 8OHdG associated with one or two copies of the six alanine repeat (ALA6) allele [88], indicating that the variant may protect DNA from ROS damage. Furthermore, the proline-leucine variant was associated with a more than twofold increase in lung cancer risk in a prospective cohort study of lung cancer [89]. The GPX1 variant could be relevant for ROS produced during cancer therapy.

3.3.4. Myeloperoxidase (*MPO*)

H_2O_2, if not neutralized, may contribute to further generation of ROS, by a reaction catalyzed by **myeloperoxidase (MPO)**. MPO generates ROS endogenously by functioning as an anti-microbial enzyme, catalyzing a reaction between H_2O_2 and chloride to generate hypochlorous acid (HOCl), a potent oxidizing agent. HOCl further reacts with other biological molecules to generate secondary radicals [90]. Thus, ultimate levels of potentially cytotoxic ROS may depend, in part, upon the balance between activities of MnSOD, CAT and MPO, determining further generation of ROS or detoxification of H_2O_2, as shown in Fig. **1**. A frequently occurring single nucleotide

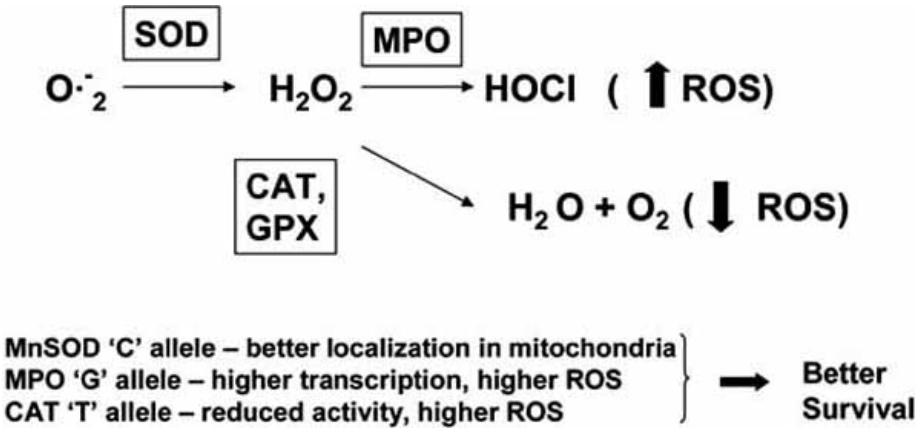

Fig. (1). Hypothesized relationships between variants in enzymes related to generation and neutralization of ROS (resulting in higher levels of ROS) and disease-free survival in patients treated with anthracyclines, alkylating agents, and taxanes. *HOCL,* hypochlorous acid; *GPX,* glutathione peroxidase.

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polymorphism in the promoter region of the MPO gene is a –463 G → A substitution, which is located in the consensus binding site of an SP1 transcription factor in the 5' upstream region of the gene [91]. The MPO A variant allele confers lower transcriptional activation than the –463 G common allele *in vitro* due to disruption of the binding site [92], and the G allele has been associated with increased MPO mRNA and protein levels in myeloid leukemia cells [93]. We found that the A allele, presumed to be associated with lower levels of ROS, was associated with decreased breast cancer risk in a large case-control study [64], and others have noted associations with decreased lung cancer risk in several, but not all studies, as reviewed in [64], as well as decreased risk of Alzheimer's disease [94]. In our breast cancer treatment and survival study, we found that high activity MPO genotypes were associated with better survival, and there appeared to be combined effects of all genotypes [74].

3.3.5. Glutathione S-transferases A1, P1, T1, M1

As reviewed by Hayes and McLellan [95], glutathione-associated metabolism is a major mechanism for cellular protection against agents that generate oxidative stress, protecting cells not only against ROS, but also against their toxic products. GSTs are induced under conditions of oxidative stress, and alpha, pi, mu, and theta-class GSTs are active in detoxification of organic epoxides, hydroperoxides, and unsaturated aldehydes, including reactive purine and pyrimidine bases and lipid peroxides produced by reactive oxidant damage to DNA and lipids, respectively [95]. GST-catalyzed reduction of these molecules prevents further oxidant damage within cells. As is the case for GPX1, GSTs are also overexpressed in many refractory tumors [96]. Thus, GST polymorphisms may influence response of cancer cells to reactive oxidant damage and to subsequent disease-free survival, as we have previously shown [61-63].

GSTA1 has a polymorphism in the 5' promoter region of the gene. Approximately 14% of a Caucasian population was homozygous for the GSTA1*B variant, and 51% heterozygous [97]. Using a reverse phase HPLC method to distinguish GSTA1 from GSTA2 protein in human liver and pancreas, Coles noted differences in the ratio of GSTA1/A2 expression associated with the GSTA1 polymorphism. Liver tissue from subjects with the GSTA1*B variant (containing 3 linked base substitutions at -52, -69, and -577 nucleotides) had decreased GSTA1 expression, and increased GSTA2 expression, compared to GSTA1*A homozygotes [97]. Directed mutagenesis for each base substitution indicated that the G → A change at position -52 was responsible for the differential promoter activities of GSTA1*A and GSTA1*B [98].

GSTP1 contains a single base substitution in exon 5 [99] that results in a variant protein with an amino acid substitution, Ile105Val, which is fairly common (51% for I105/I105, 43% for I105/V105, 6% for V105/V105 in Caucasians) [99]. Human GSTM1 and GSTT1, each has a gene deletion polymorphism, resulting in loss of enzyme expression. Homozygous deletion of *GSTM1* and *GSTT1* occurs in approximately 50% and 18% of Caucasian populations, respectively [100]. Individuals lacking these enzymes may have reduced removal of oxidation products produced by cancer therapy and thus, better prognosis. Indeed, we have previously found that variants in all the GSTs resulting in decreased (GSTA1, GSTP1) or absent (GSTM1, GSTT1) enzyme activity were associated with significantly improved survival among women receiving treatment for breast cancer [61-63]. More recently, associations for *GSTP1* and breast cancer survival were corroborated in the Shanghai Breast Cancer Study [101].

CONCLUSION

Although use of antioxidant supplements is widespread among cancer patients, the impact of supplement use on treatment outcomes remains a controversial area. There are limited data to guide physicians in providing recommendations to their patients regarding use of antioxidant supplements during treatment. This area merits further investigations, based on the recognition that a major mechanism for the cytotoxic activity of numerous chemotherapeutic agents is through increased formation of ROS and apoptosis, and intriguing evidence that antioxidants

may potentiate the effects of chemotherapeutic agents, with different effects in normal *vs*. tumor cells. Although it may be premature to randomize patients to antioxidants during chemotherapy in a clinical trial due to the potential for negative effects on therapeutic efficacy, large well-designed observational studies are needed to provide data that can guide therapeutic decision-making, as well as provide information for potential clinical trials of antioxidants during treatment.

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